Strategies for Ensuring Potency of Adenovirus-Based Vaccines

Katey Einterz Owen, PhD
Merck Research Laboratories
11 October 2005

MERCK
Research Laboratories

Merck's Candidate HIV Vaccines

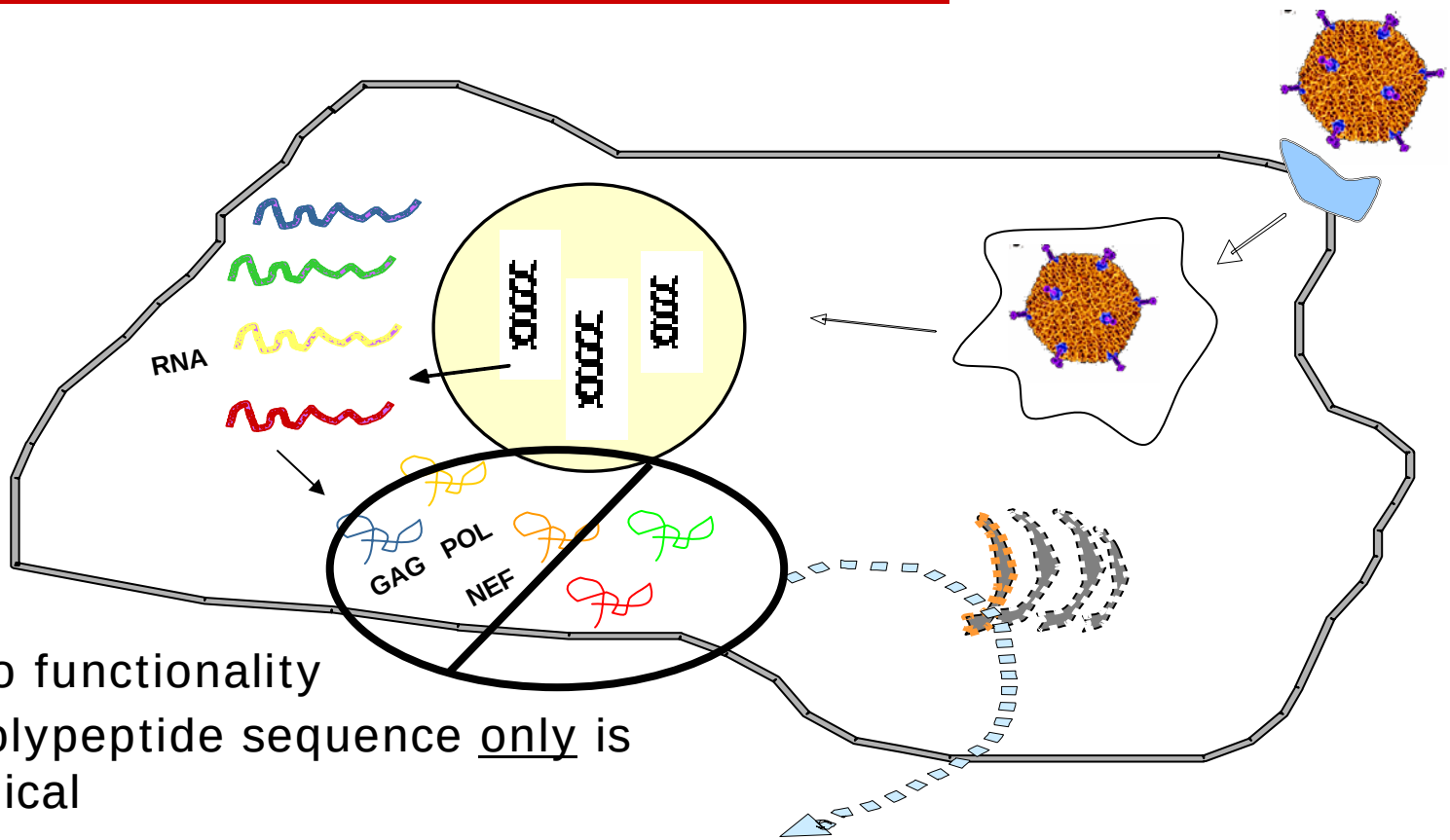
- MRKAd5gag
  - Has been in numerous PhI trials, including dose ranging
- MRKAd5trivalent (gag+pol+nef)
  - PhI dose ranging complete
  - PhII POC study initiated Dec. 2004
- MRKAd5 nef/gag/pol/ + MRKAd6 nef/gag/pol (trigene)
  - An alternate approach
- Other constructs as required

Merck's Adenovirus-Based Vaccines

- Other Adeno-based vaccines are also in development for various indications, at various stages

- Hoping to develop a single, coherent release strategy for all Adenovirus-based vaccines
  - Clear distinction between vaccines and gene therapy indications

Mechanism of Vaccine Activity



- No functionality
- Polypeptide sequence only is critical

Presented by MHC to T-Cells → Immune Response

MERCK Research Laboratories

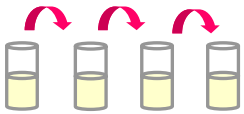
The Charge to Analytics

- Ensure the vaccine is safe and efficacious, as determined clinically

- Best way to accomplish this:
  - Make sure vaccine for launch is equivalent to vaccine in trials

Dose—
The Genome Quantitation Assay

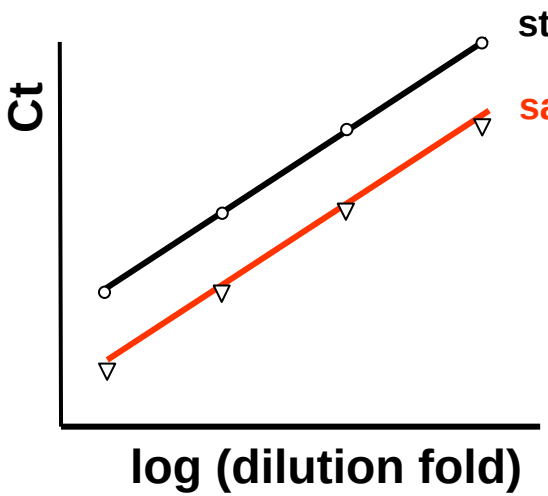
Dilute Samples



Lysis
SDS & Heat

+ PCR master mix

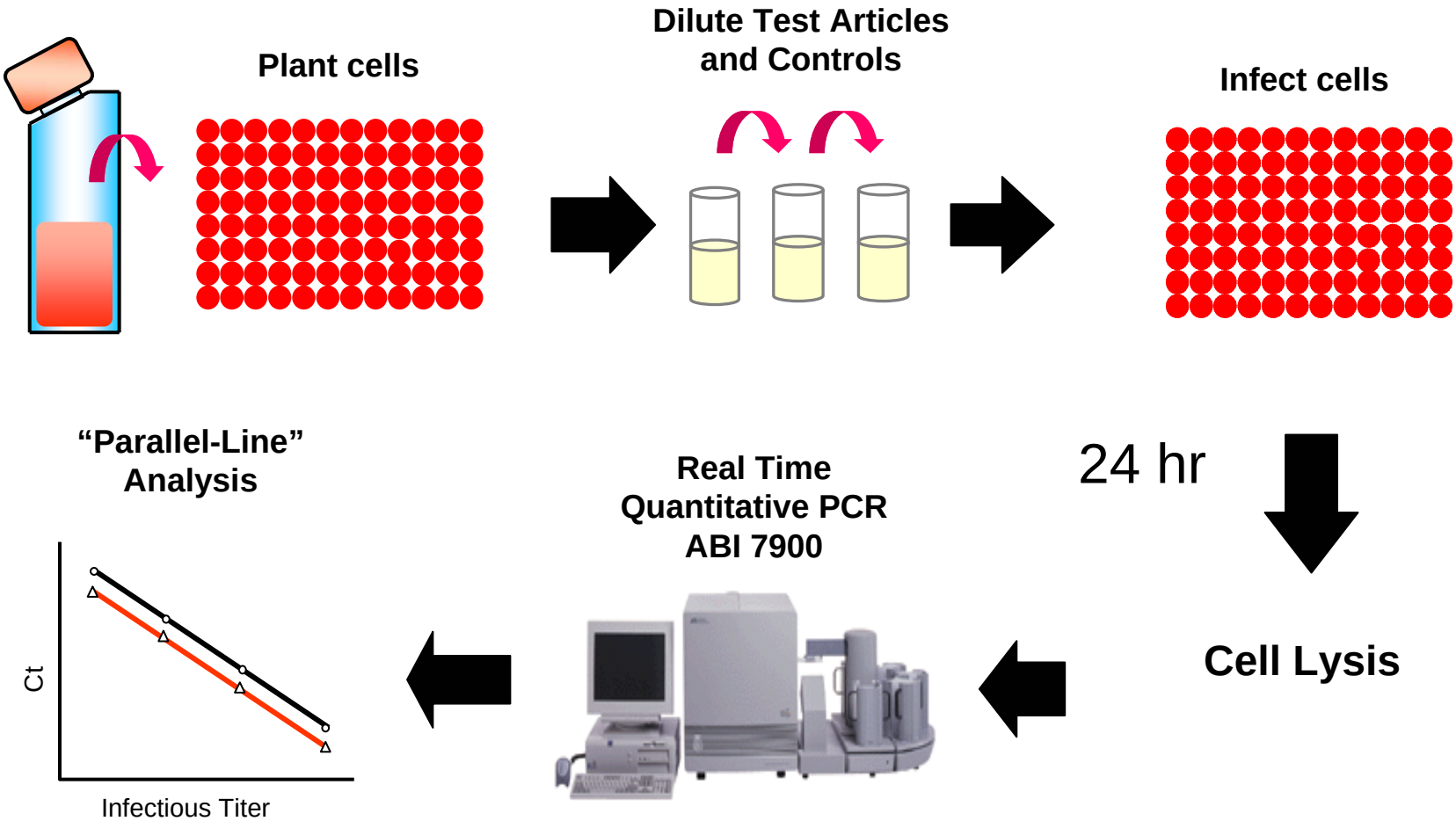
Real-time QPCR



Summary of the GQA

- Variance component anaysis (VCA) suggests ~5-10% root variability
  - Encompasses variability across operators and across laboratories
- Accurate
  - Good correlation between serotype-specific and additive transgene-specific result
- Specific
  - Clearly distinguishes various components
- NOT stability indicating

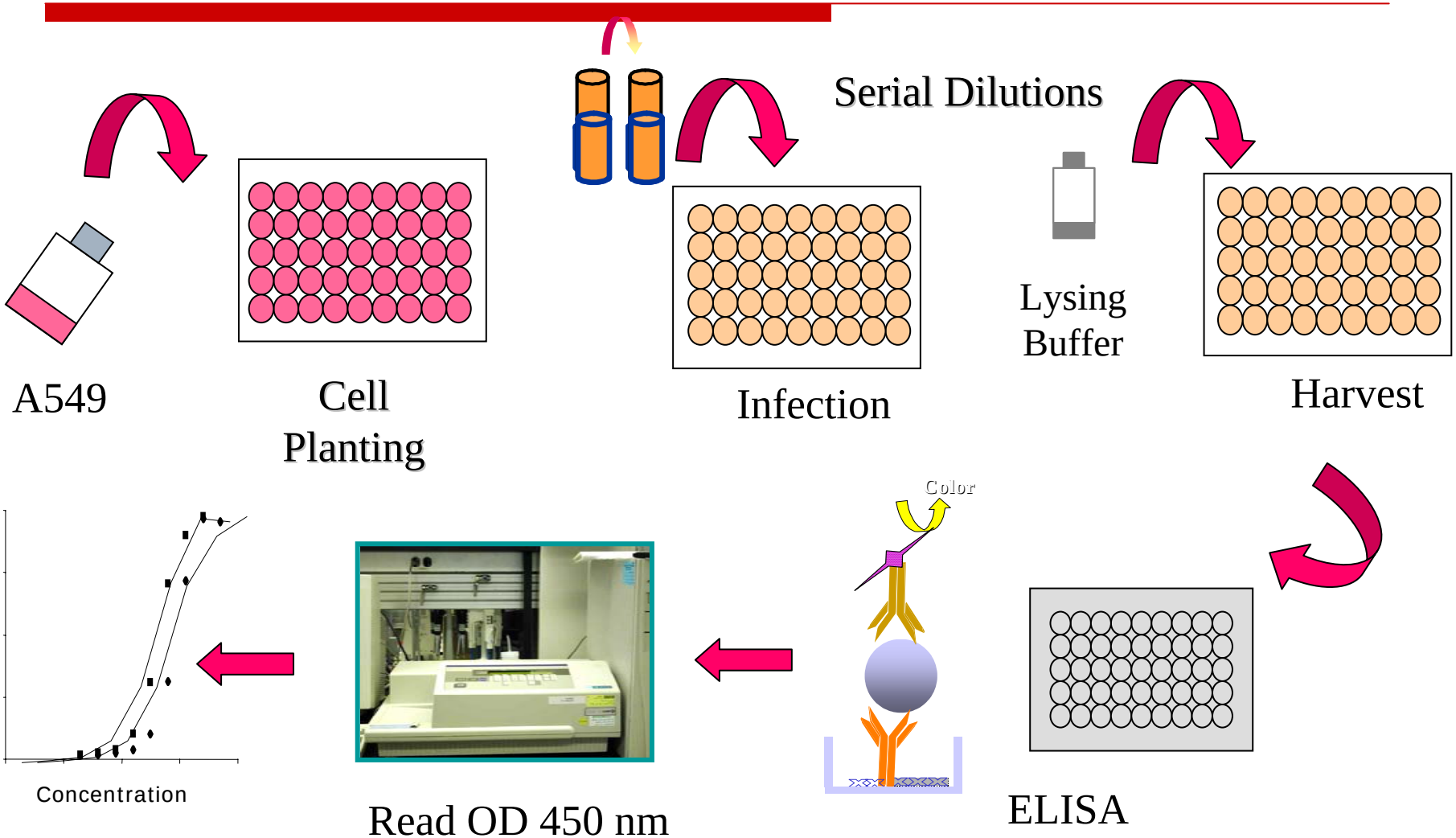
Potency—The Q PCR-Based Potency Assay (QPA) for Infectivity



Summary of the QPA

- Variance Component Analysis (VCA) suggests ~20-25% root variability
  - Encompasses variability across operators and across laboratories
- Accurate
  - Good correlation between serotype-specific and additive transgene-specific result
- Specific
  - Clearly distinguishes various components
- Stability Indicating

In Vitro Antigen Expression Assay

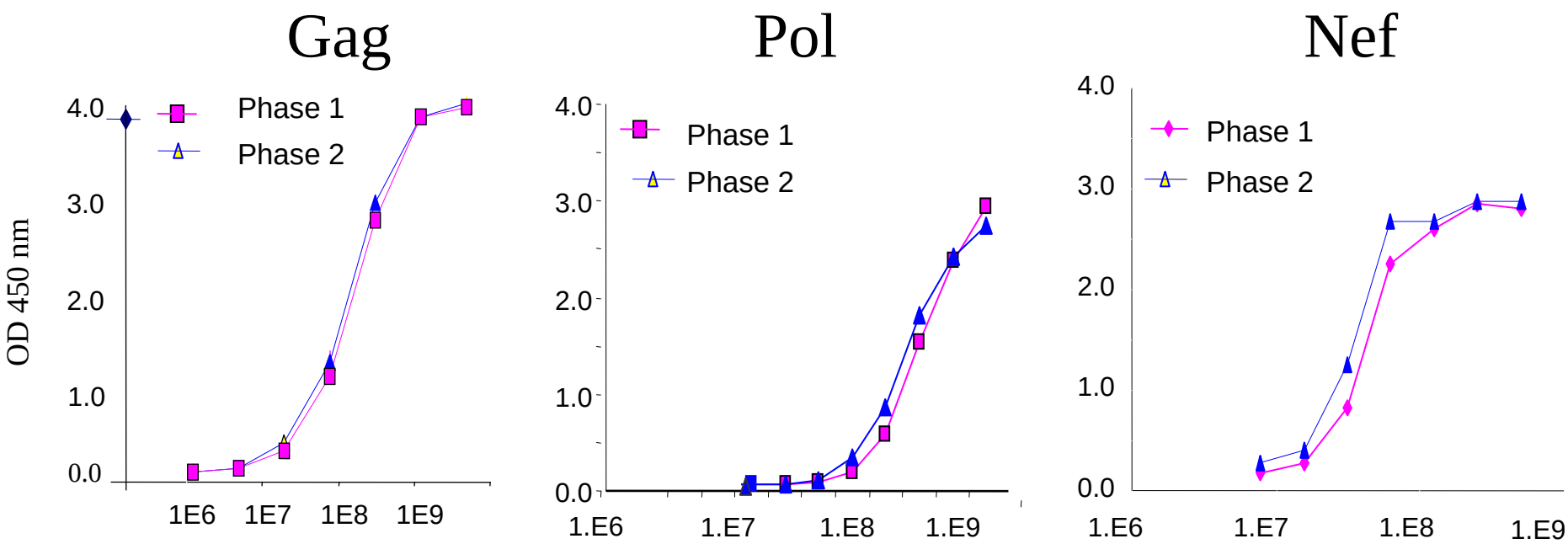


J. Rolling, S. Zhang, M. Geary, D. Hsu

MERCK Research Laboratories

Relative Potency of Monovalent Bulks by IVAE

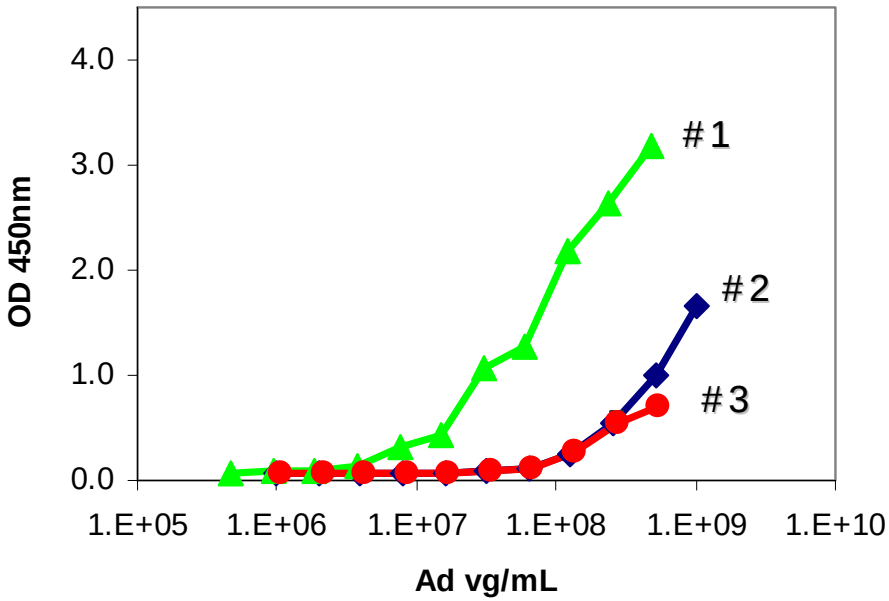
(J. Rolling, S. Zhang, M. Geary, P. Tsai)

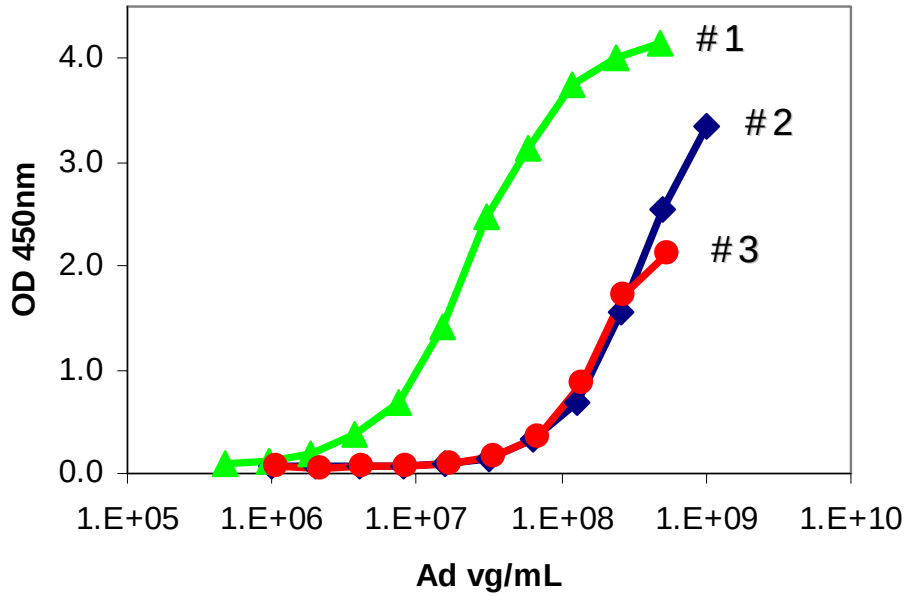


- Results suggest essentially equivalent expression for PhI/PhII bulks
- These are <u>not</u> typically representative results, but “the best” results we’ve seen
- Assay is not optimized (no “top of the curve” for pol; ODs > 2.0)

MERCK Research Laboratories

More Typical IVAE Results

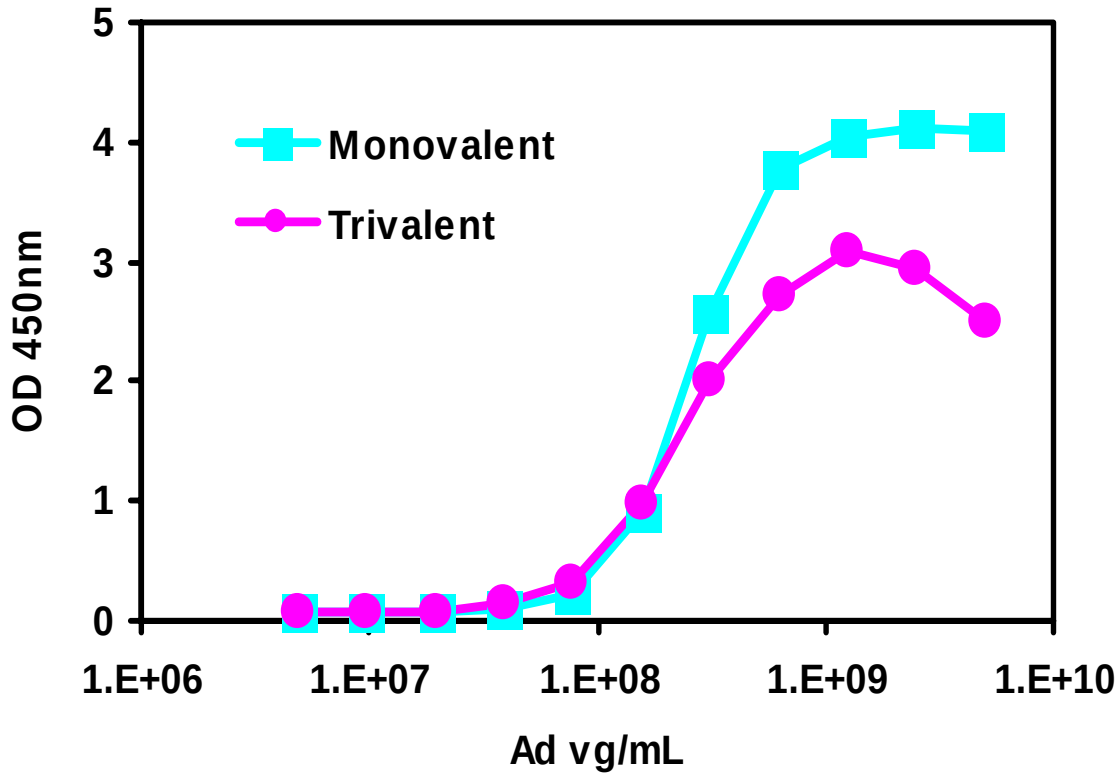




- Assay is not robust in its current state
- Considerable efforts in development have improved robustness, but not yet sufficiently

More Problems--Interference in Expression in Multivalent Samples

J. Rolling, M.N. Takahashi

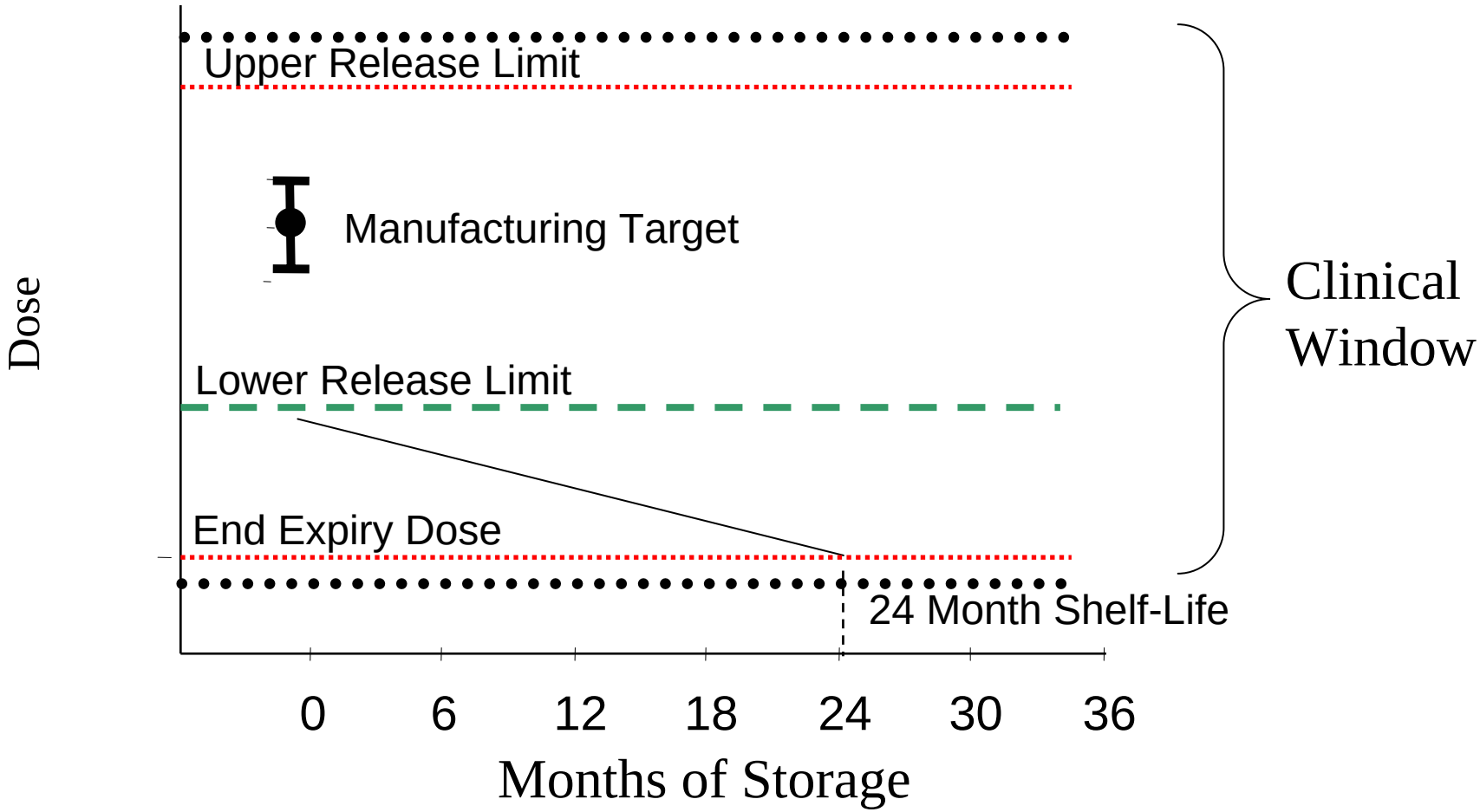


- Data indicate that pol and nef in the monovalent vaccine are more highly detected than in the trivalent vaccine—clinical relevance unclear
- Raises issue of whether or not an appropriately-relevant standard could ever be generated for a trivalent vaccine

Summary of the IVAE

- Variability is high
  - No VCA yet, but assay is influenced by factors we don't yet understand
- Not necessarily accurate
  - Poor correlation between monovalent and trivalent expression
  - Clinical relevance of interference is unclear
- Specific
  - Clearly distinguishes various components
- Probably is stability-indicating

Vaccine Fill and Release "Model"



Merck's Proposed Analytical Strategy

- Demonstrate effective potency clinically through end-expiry studies
- Measure clinical materials for total full particles (genome content), in vitro infectivity (qPCR-based), and in vitro antigen expression
- Measure Process Validation lots for all three attributes

Merck's Proposed Analytical Strategy

- Assuming clinical success and correlation with analytical methods, we propose to measure only total particles and infectious particles for routine release

- Approach assumes:
  - Correlation between total particles and clinical safety
  - Correlation between in vitro infectivity and clinical efficacy
  - Reasonable correlation between in vitro infectivity and in vitro antigen expression
  - Demonstrated genetic stability of the construct

Summary—Currently Proposed Strategy

- A measure of total full particles will define "dose"
- Infectivity, measured using qPCR, will define "potency"
- Both dose and potency will have independent, clinically-based specifications
  - Currently no other release or stability assays for dose or potency of filled containers will be performed, including IVAE

Acknowledgments

GQA

Chris Wang
Liman Wang
Jenny Xu
Diana Pacchione

QPA

Liman Wang
Jayanthi Wolf
Danielle McElhaugh
Jennifer Naspinski

IVAE

Judy Rolling
Marie-Noëlle Takahashi
Robyn Reinmiller
Margie Geary
Shawn Zhang

Assay Support

Jennifer McMackin
Heather Baxter
Laura McCoach
Scott Barmat

John Lewis
David Hsu
Pei-Kuo Tsai
John Hennessey

The Merck Vision

"We try never to forget that medicine is for the people. It is not for the profits. The profits follow, and if we have remembered that, they have never failed to appear."

--George W. Merck
December 1950